Material selection and preparation process: challenges and outcomes

G. Roebben

Joint Research Centre

Institute for Reference Materials and Measurements

(with contributions from LGC, BAM, NPL, PTB, HAS)

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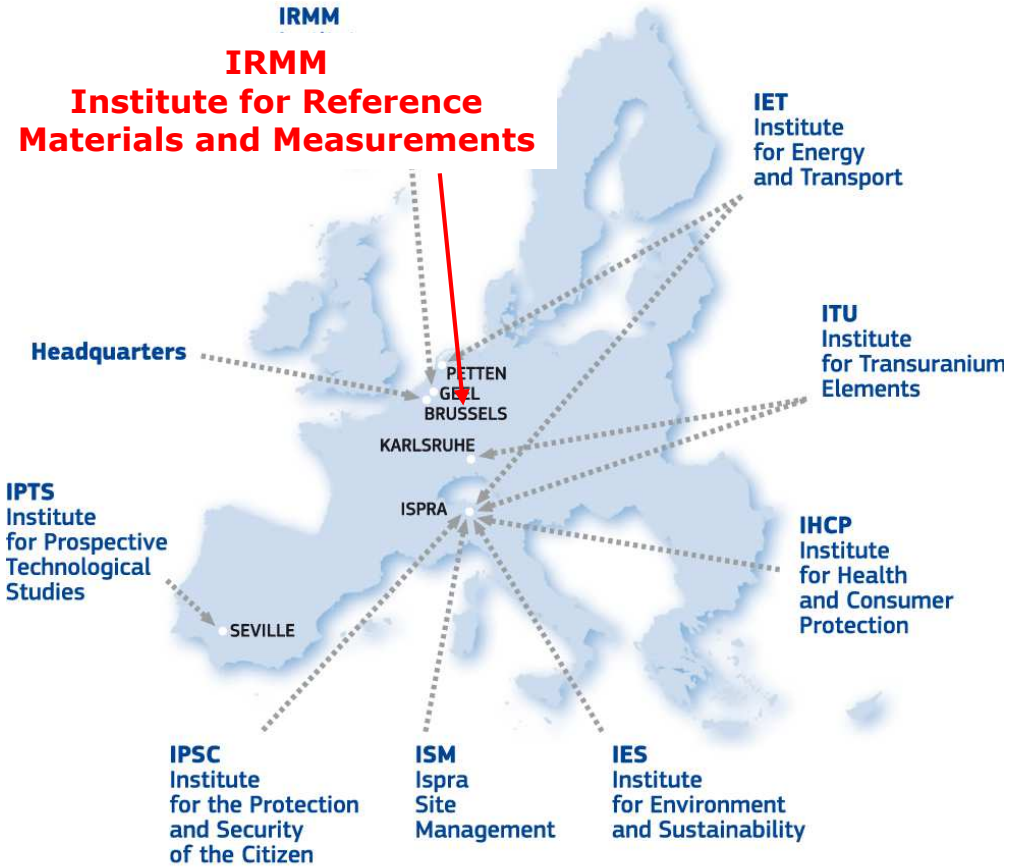
The Joint Research Centre (JRC)



JRC

- European Commission's in-house science service
- Supporting EU policies with independent, evidence-based scientific and technical advice
- ~ 3.000 staff, 6 locations

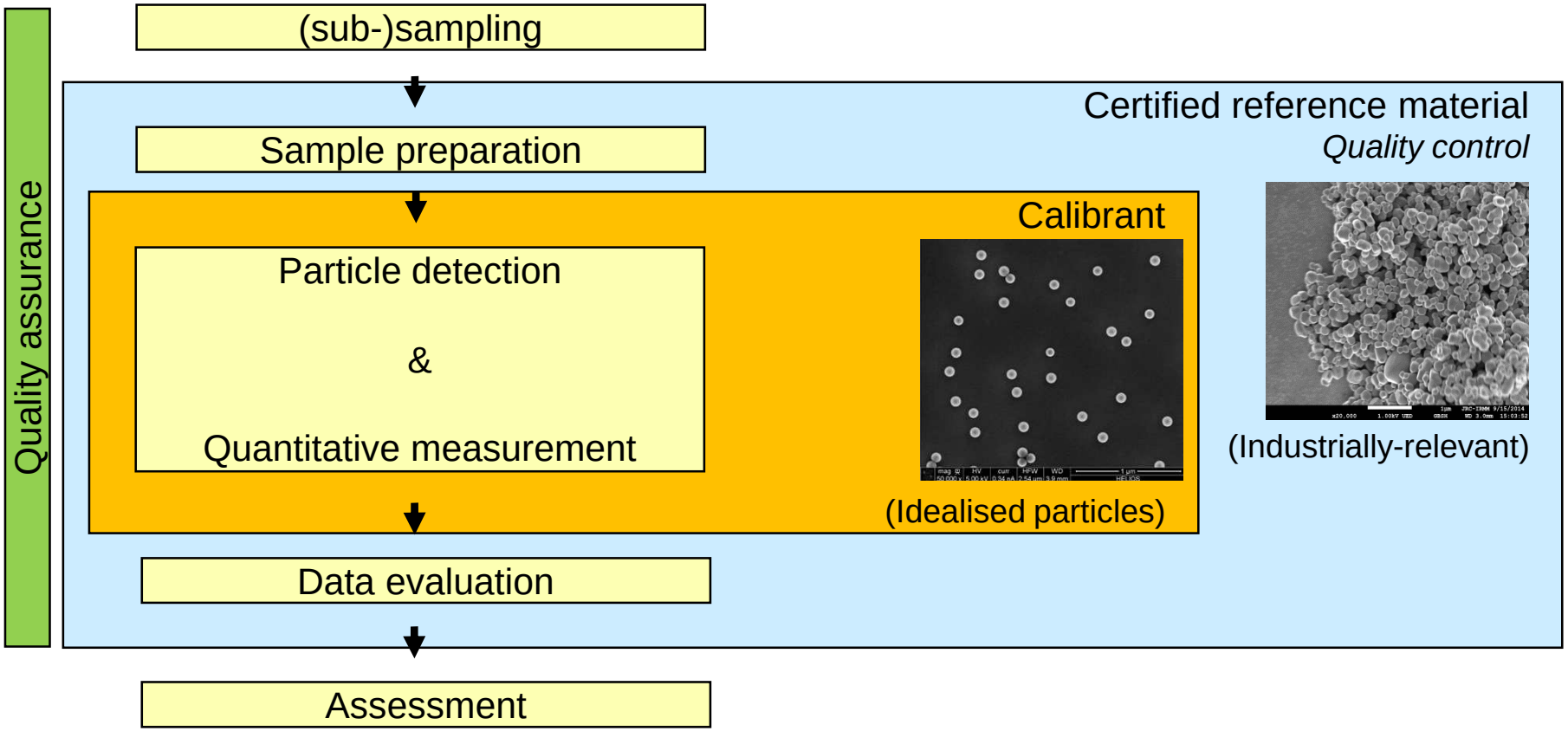
IRMM (Geel, Belgium)

- Confidence in measurements®
- ~ 250 staff
- 4 Scientific Units

JRC-IRMM's reference materials

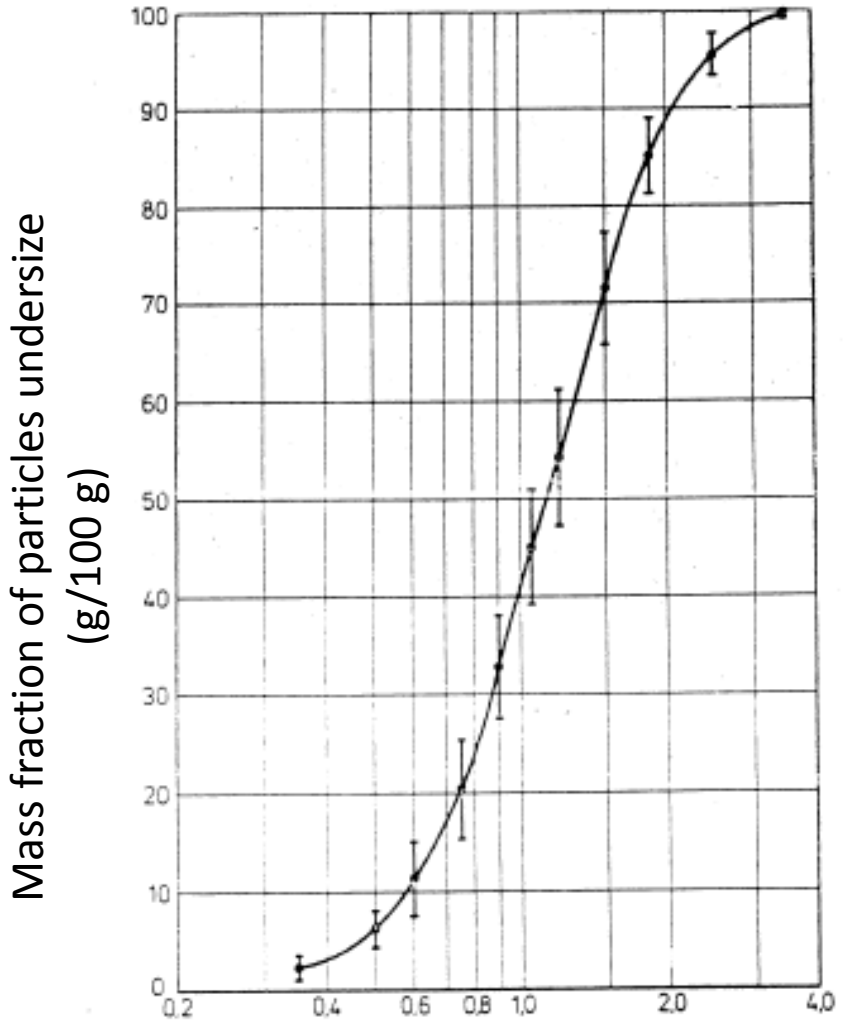
- JRC-IRMM is a leading reference material developer worldwide
- Materials range from nuclear, industrial, food, GMOs, environmental to clinical materials
- 780 different materials available (measurement standards)
- 20,000 units distributed yearly; 20-30 new materials yearly

Role of RMs in reliable particle size analysis



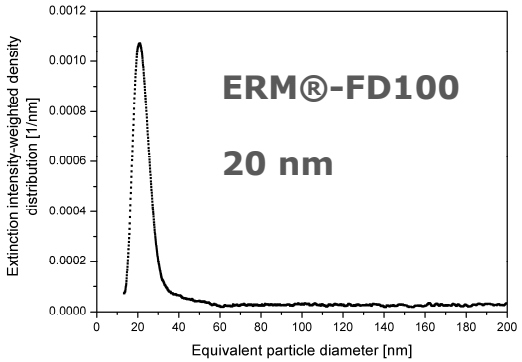
IRMM particle RMs?

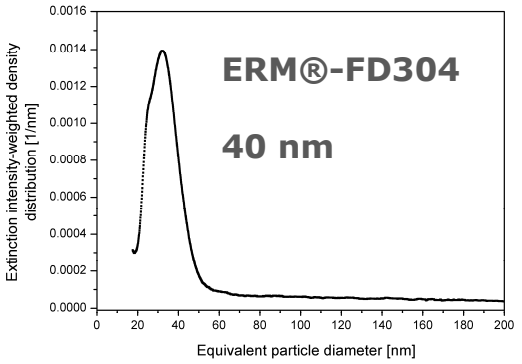
Wilson et al., EUR 6825 (1980)



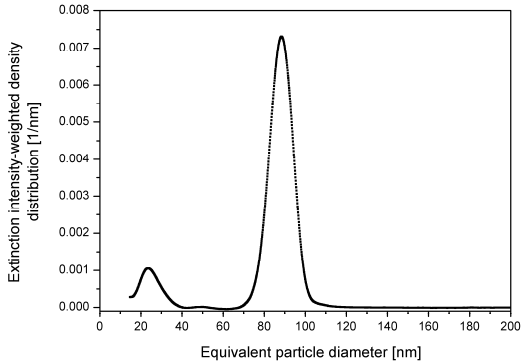
European Commission

Silica nanoparticles with monomodal particle size distributions





Silica nanoparticles with bimodal particle size distribution



ERM®-FD102

Nominal particle size 20 nm and 80 nm

Silica and other industrially relevant nanoparticles:

- **Non-spherical**
- **In a complex matrix**
- **With a polydisperse size distribution**

Increasing complexity

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NanoChOp

Why reference materials for NanoChOp?

- Common test materials:
  enable broader characterisation via combining data from multiple partners:
  - size distributions
  - particle concentrations
  - microbiology tests
  - impurities

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  - particle concentrations
  - microbiology tests
  - impurities

- Reference materials: improve understanding of variance within and between laboratories and methods:
  - Quantified heterogeneity
  - Quantified stability

What is a reference material?

Reference material (RM):

material, sufficiently homogeneous and stable with respect to one or more specified properties, which has been established to be fit for its intended use in a measurement process

ISO Guide 30:2015(en), 2.1.1

What is the role of RMs in NanoChOp?

Intended use:

… nanoparticle reference materials composed of oxide materials (to develop methods for physical and chemical characterisation), fluorescently labelled oxide materials (to allow nanomaterials tracking within biological systems to be performed) and a quantum dot nanomaterial (to develop methods for the optical characterisation of fluorescent nanomaterials)…

Agreed target properties for RMs (1)

Material properties	Target values/characteristics		
	SiO_2	TiO_2	Quantum dots
Size (nominal particle diameter)	50 nm	20 nm	30 nm
Polydispersity (FWHM/mean diameter)	< 0.25	< 0.25	< 0.25

Model materials:

- spherical, essentially monodisperse
- to match with complex task of measuring in serum

Agreed target properties for RMs (2)

Material properties	Target values/characteristics		
	SiO_2	TiO_2	Quantum dots
Size (nominal particle diameter)	50 nm	20 nm	30 nm
Polydispersity (FWHM/mean diameter)	< 0.25	< 0.25	< 0.25
Surface functionalization	Aminated	None	Aminated or carboxylated
Zeta potential (absolute value)	> 10 mV	> 10 mV	> 30 mV
Mass fraction	> 5 g/kg	> 5 g/kg	about 20 g/kg
Amount (mass of particles)	12 g	7 g	7 g
Shelf-life (in closed containers)	18 months	18 months	18 months
Shelf-life (after opening containers)	5 days	5 days	5 days

	NanoChOp-01	NanoChOp-02	NanoChOp-03	NanoChOp-04	NanoChOp-05	NanoChOp-06
Core particles	**silica** (with organic fraction)	**silica**	**CdSe / CdS / ZnS**	**silica** (with organic fraction)	**silica**	**silica**
Surface groups	$-NH_2$	None	amine – PEG	-COOH	none	$-NH_2$
Amount	**523 units** 2.5 mL/unit 2.5 g/kg	**540 units** 2.5 mL/unit 2.5 g/kg	**228 units** 2.5 mL/unit ~ 4 g/kg	(not ampouled)	**400 units** 9 mL/unit 2.5 g/kg	**536 units** 1.9 mL/unit 2.5 g/kg
Polydisper-sity (based on SAXS)	0.10	0.15	(-)	0.11	0.13	0.13

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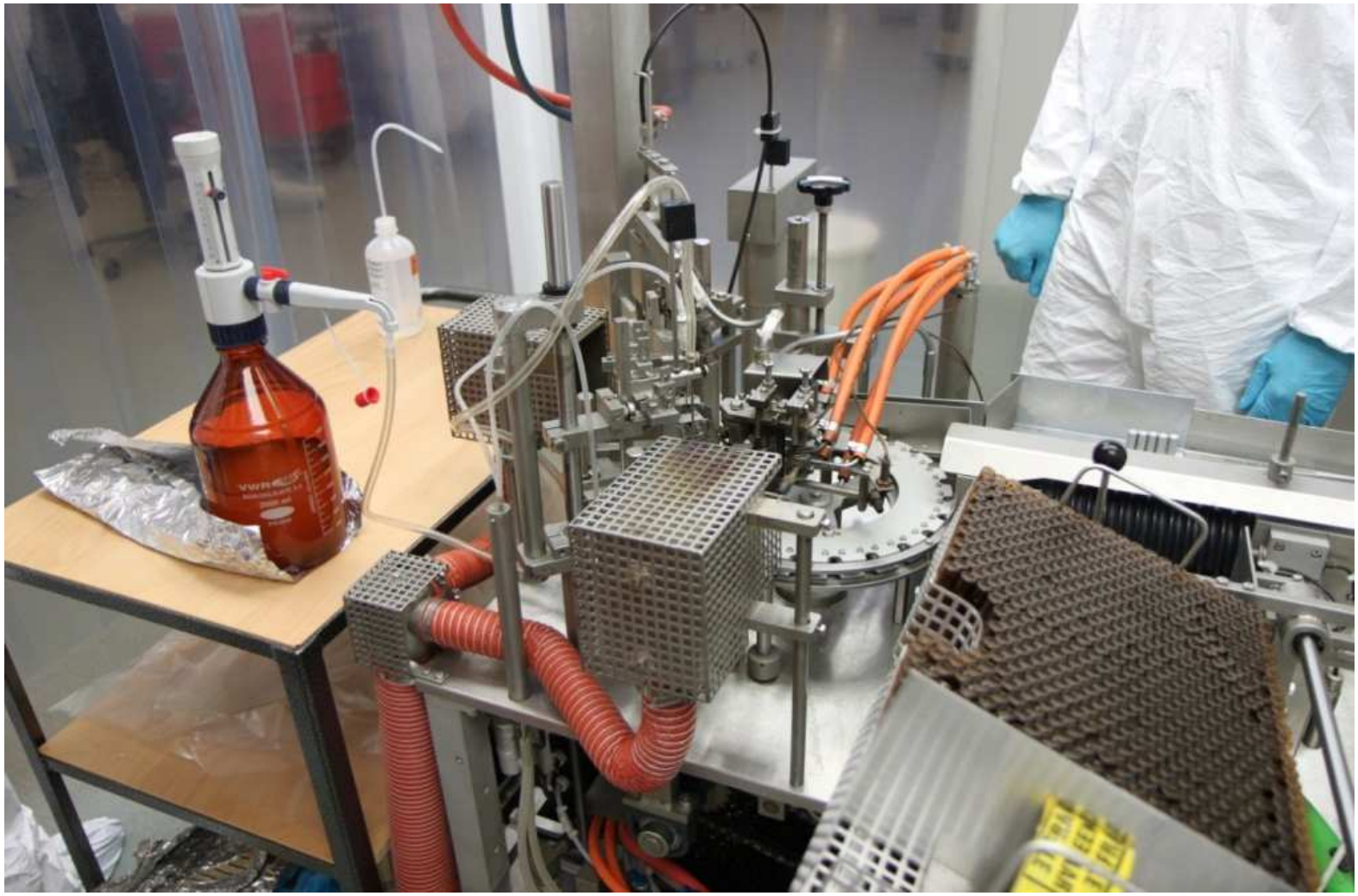
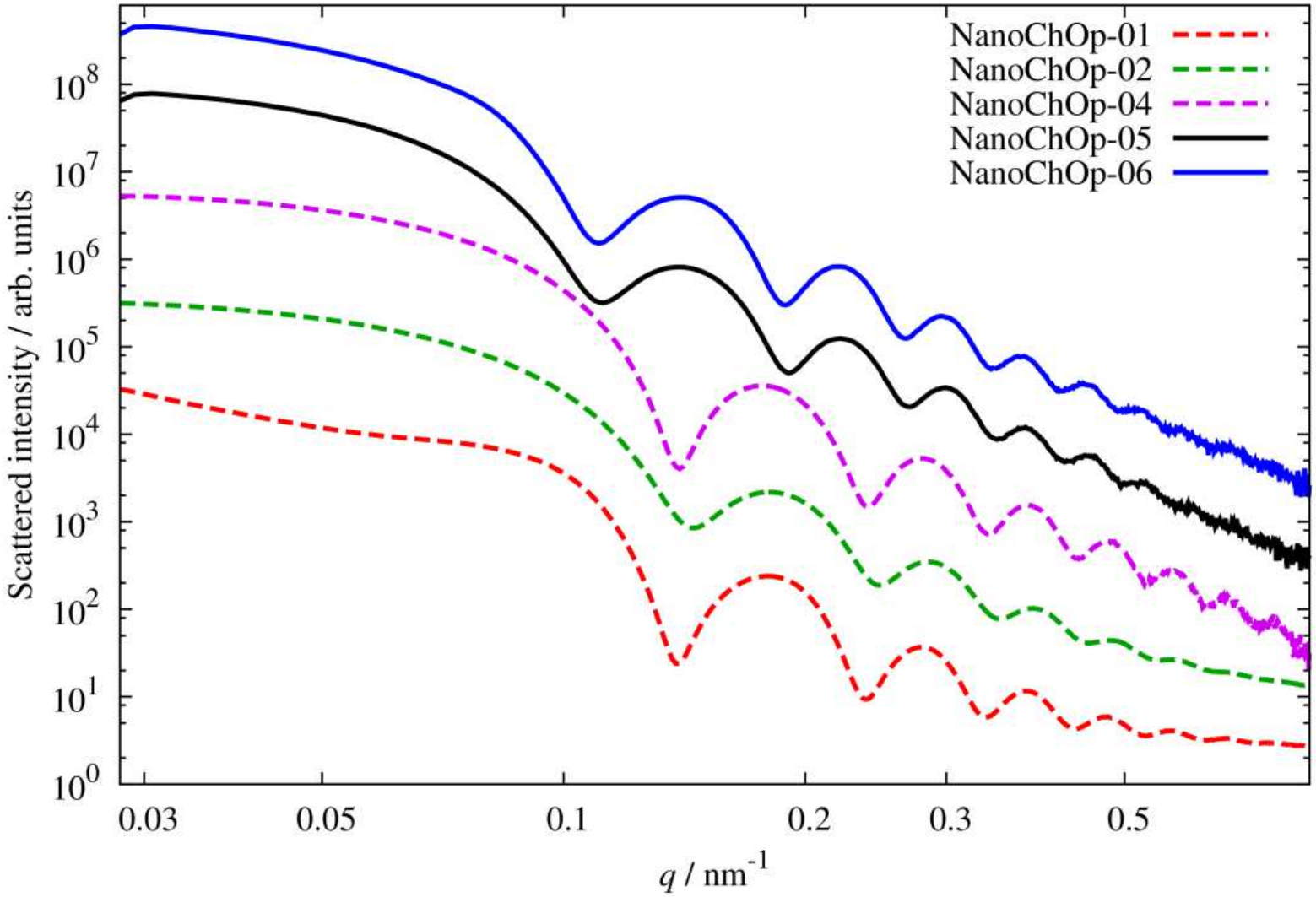
Polydisper-sity (based on SAXS)	0.10	0.15	(-)	0.11	0.13	0.13
Remedia-tion of microbio-logical contami-nation	**gamma irradiation** (part of batch)	**autoclaving** (part of batch)	**gamma irradiation**	-	long term storage in air-tight closed glass ampoules	not required

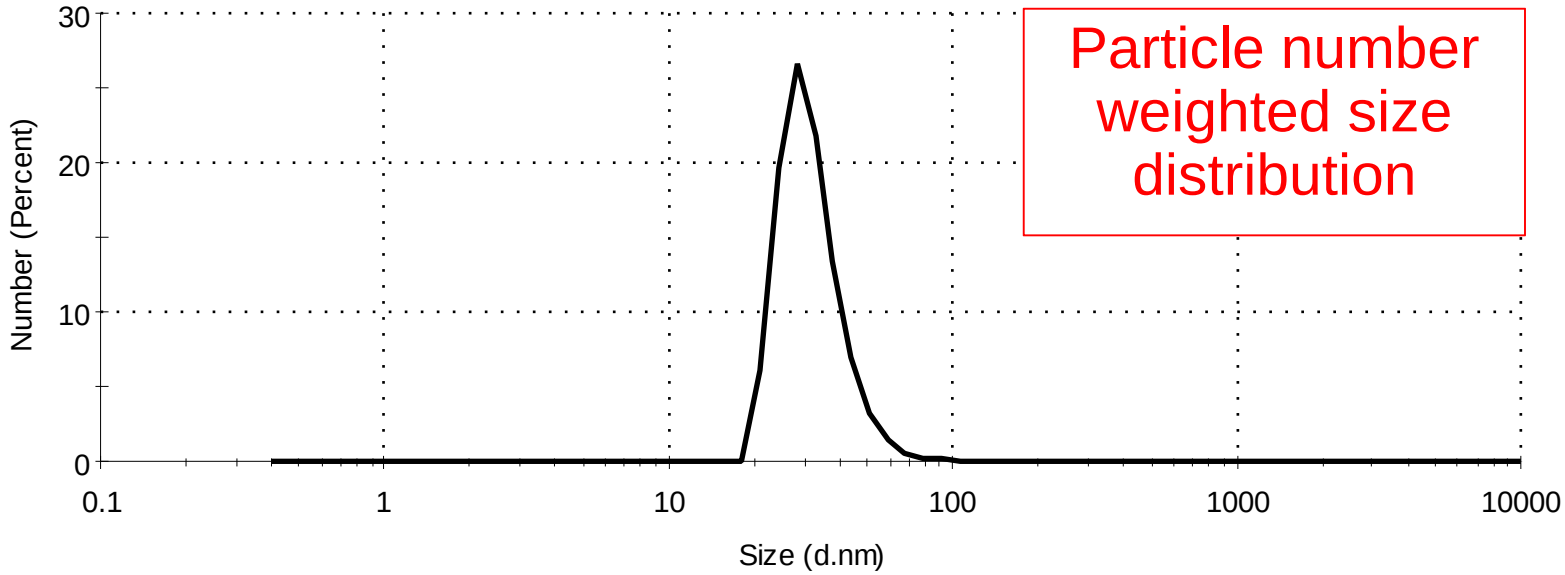
Ampouling nanoparticle suspensions in mobile clean cell

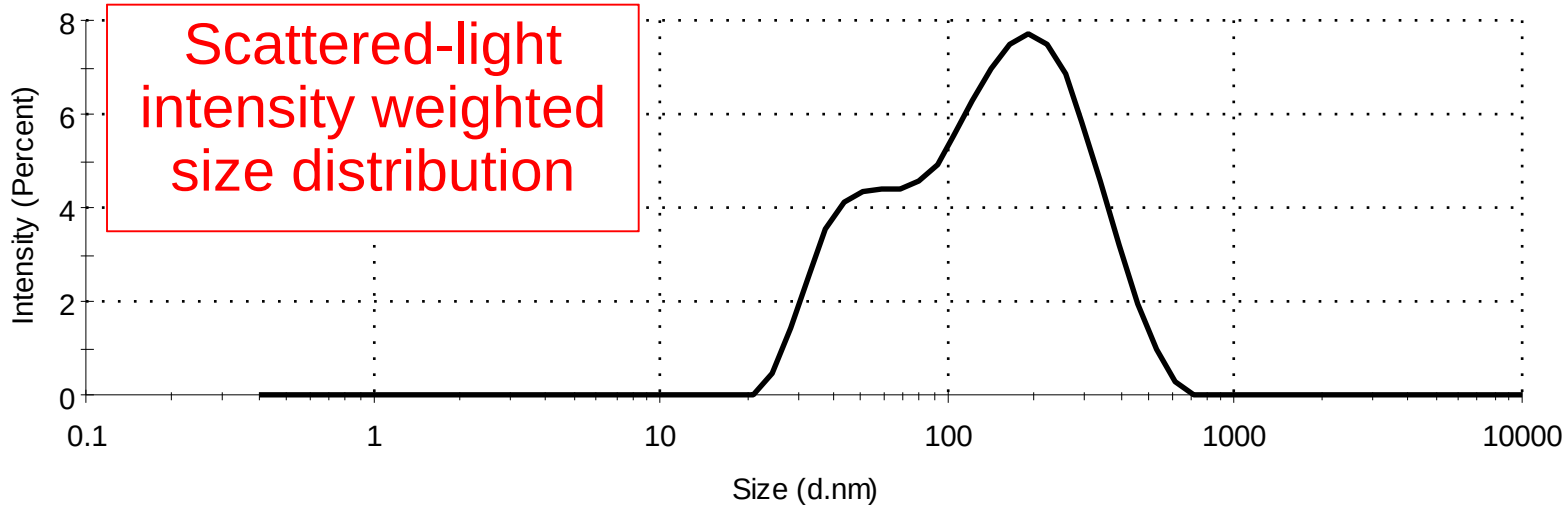


SAXS curves for NanoChOp silicas



NanoChOp-03 QDs: DLS size distributions





Between-unit homogeneity assessment

Material	Measurand	Average value	Homogeneity (standard uncertainty, u_h)
NanoChOp-03	$d_{DLS,cum}$	31 nm	**7 %**
	$d_{DLS,NNLS,nb}$	103.2 nm	2.2 %
	ζ_{ELS}	-1.4 mV	0.6 mV

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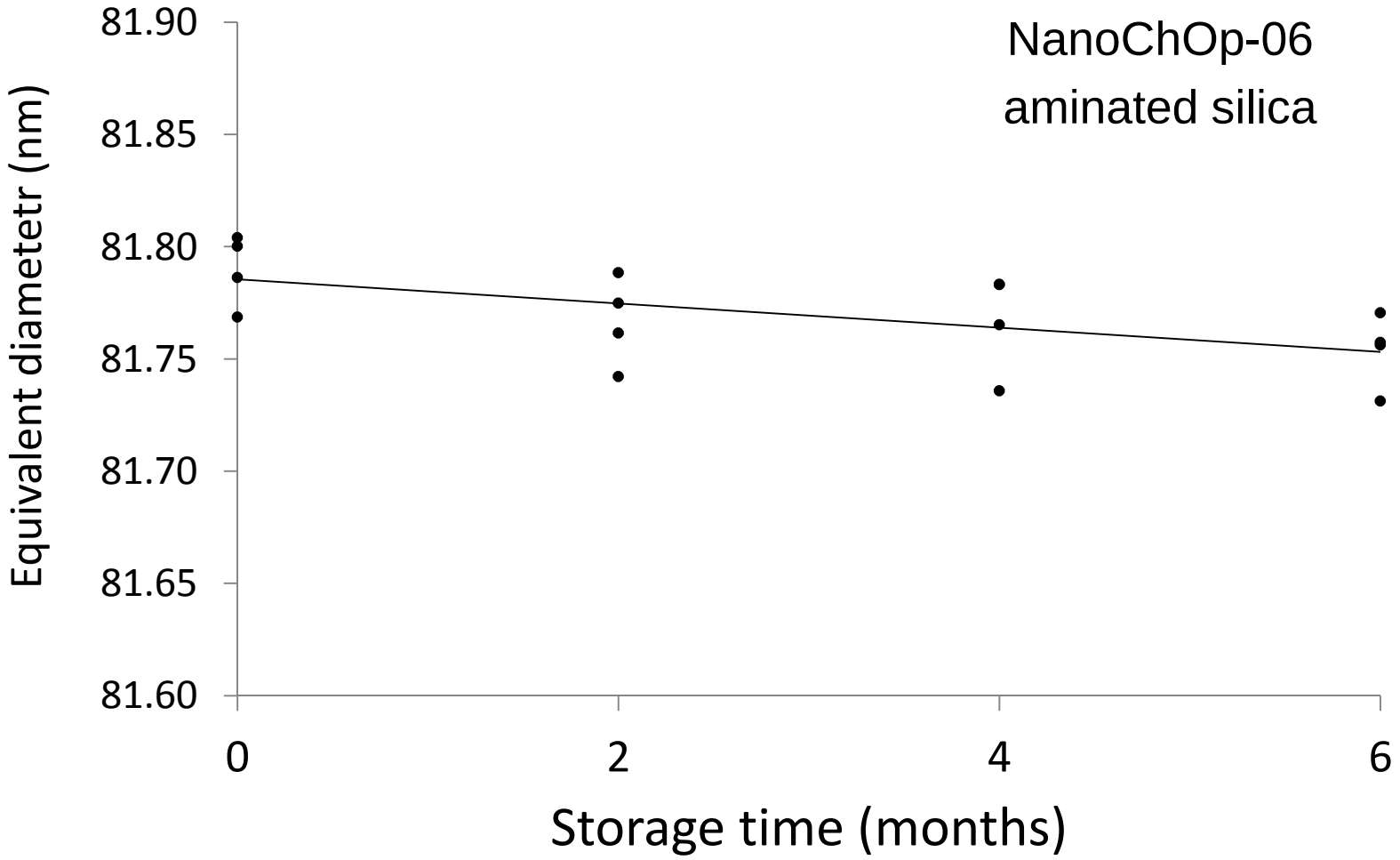
	$d_{DLS,NNLS,nb}$	103.2 nm	2.2 %
	ζ_{ELS}	-1.4 mV	0.6 mV
NanoChOp-05 (silica)	$d_{DLS,NNLS,I}$	94.3 nm	1.0 %
	$d_{CLS,I}$	86.9 nm	0.5 %
	ζ_{ELS}	-48.3 mV	1.8 mV

Between-unit homogeneity assessment

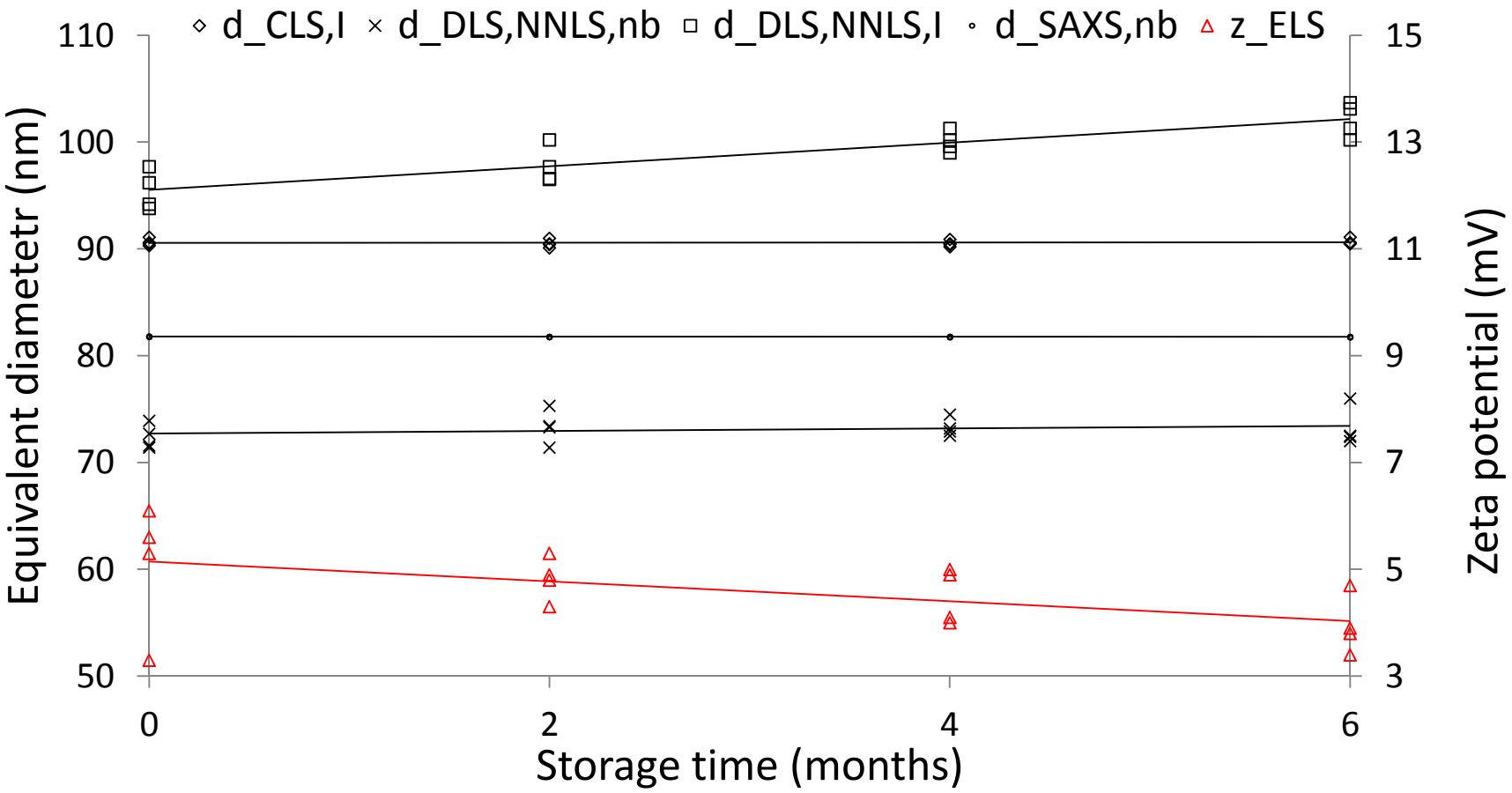
Material	Measurand	Average value	Homogeneity (standard uncertainty, u_h)
NanoChOp-03 (QDs)	$d_{DLS,cum}$	31 nm	**7 %**
	$d_{DLS,NNLS,nb}$	103.2 nm	2.2 %
	ζ_{ELS}	-1.4 mV	0.6 mV
NanoChOp-05 (silica)	$d_{DLS,NNLS,I}$	94.3 nm	1.0 %
	$d_{CLS,I}$	86.9 nm	0.5 %
	ζ_{ELS}	-48.3 mV	1.8 mV
NanoChOp-06 (aminated silica)	$d_{DLS,NNLS,I}$	89.9 nm	0.3 %
	$d_{CLS,I}$	88.4 nm	0.2 %
	$d_{SAXS,nb}$	81.8 nm	0.02 %
	ζ_{ELS}	9.7 mV	0.8 mV

Stability assessments (1)



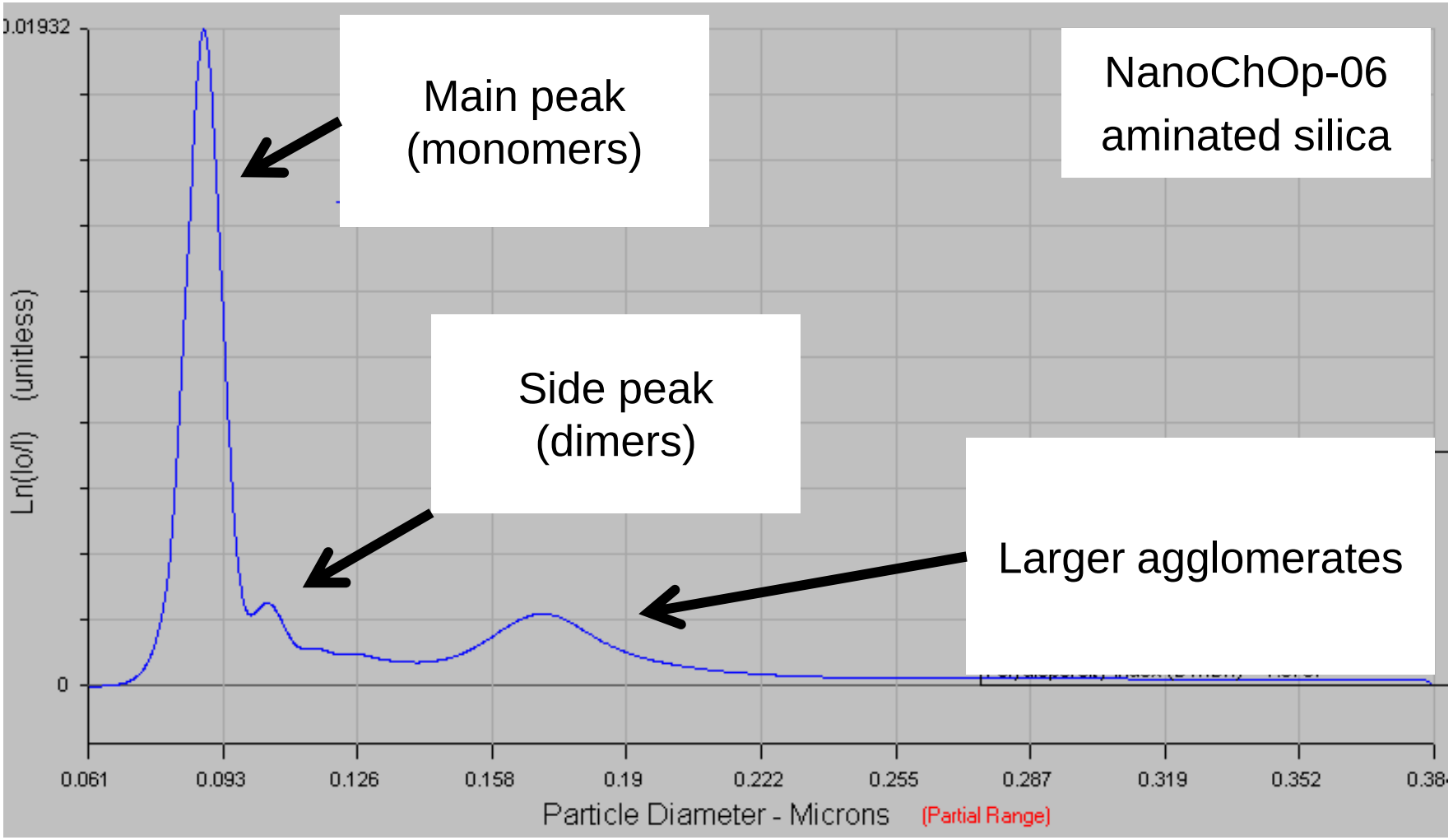
Long term stability study (18 ^{o}C) via Small-angle X-ray scattering, $d_{SAXS,nb}$

Stability assessments (2)



NanoChOp-06 stability study (18 ^{o}C) via SAXS, CLS, DLS, ELS

Stability assessments (3)



Size distribution via (Disc) centrifugal liquid sedimentation, $d_{CLS,i}$

Material information sheets

	NanoChOp-05			
General description	silica nanoparticles, nominal mass fraction 2.5 g/kg			
Specific observations	Free of active bacterial contamination			
Instructions for use	Remove cloud formation by repeated inversion; minimum sample intake 0.3 mL; store at room temperature; avoid freezing; close opened ampoule with paraffin film; use within 10 days after opening (or on day of opening for measurement of ζ_{ELS}).			
Equivalent diameters	**Measured value**	u_h	U_{lts} (36 months)	**status**
$d_{DLS,cum}$	90 nm	-	1.1 %	**RM**
$d_{DLS,NNLS,i}$	94 nm	1.0 %	2 %	**RM**
$d_{CLS,i}$	87 nm	0.5 %	2 %	**RM**
$d_{SAXS,nb}$	(81.1 ± 0.8) nm	-	-	**RM**
Other measured properties				
ζ_{ELS}	-48 mV	1.8 mV	-	**RTM**
pH	8.4	-	-	**Test material**
Effective particle density	2.0 g/cm^3	-	-	**Test material**

What is a representative test material?

Representative test material (RTM):

material, which is sufficiently homogenous and stable with respect to one or more specified properties, and is implicitly assumed to be *fit for its intended use in the development of measurement and test methods that target properties other than those for which homogeneity and stability have been demonstrated*

ISO/TS 16195:2013(en), 3.1

Summary

- 3 materials were made available as common test materials, with material information sheets.
- Stability and homogeneity (and reference material status) depend on relevant size parameter.
- Improved understanding of capacity of different size analysis methods to assess homogeneity and stability of nanoparticle suspensions.
- Increased know-how on preparing batches of sterile nanoparticle suspensions as common test materials for use in biologically relevant media.

Other WP1 results

1. Selection of common medium (EMEM), serum (FBS) and cell line (HepG2) (LGC)
2. Fluorescent labeling of aminated silica (BAM)
3. Agreed dispersion protocols (LGC)

Impact

- Actual impact:
  - availability nanoparticle reference materials (silica and aminated silica) or test materials (quantum dots) for use in other NanoChOp work packages and beyond.

- Expected future impact:
  - availability of certified nanoparticle reference materials for use in more complex (biological) media.

Acknowledgements

- V. Kestens, Y. Ramaye, T. Gerganova, …
  - (JRC-IRMM Engineering Materials Laboratory)
- J. Charoud-Got, …
  - (+ JRC-IRMM Reference Material Processing Group)
- Collaborators in our CRM characterisation studies
- The funding of the EU through EMRP

And: our partners in NanoChOp!

European Commission

Training course: use of reference materials and the estimation of measurement uncertainty

Date: 7-8 October 2015

Location: IRMM (Geel, Belgium)

Deadline for registration: 7 September 2015

Max. number of participants: 30

Info: jrc-irmm-rm-course@ec.europa.eu

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